

CORRECTED VERSION OF THE CLAIMS

A2 1. (Amended) A reversible physiological process for temporal separation of oxygen evolution to avoid deactivation of hydrogenase in the presence of oxygen and ^{to} sustain photosynthetic hydrogen production in cells of an algal microorganism, comprising:

(a) growing a culture of the cells of the algal microorganism in a medium under illuminated conditions to accumulate an endogenous substrate;

Sub B (b) depleting a nutrient selected from the group consisting of sulfur, iron, and/or manganese from the medium;

(e) ^{c?} sealing the culture from atmospheric oxygen;

(f) ^{d?} incubating the culture in light to provide a rate of light-induced oxygen production equal to or less than a rate of cellular respiration; and

(e) collecting an evolved gas that includes hydrogen.

But D2 2. (Amended) The process of claim 1 wherein said hydrogen gas is generated from water and said endogenous substrate using light and a hydrogenase.

3. (Amended) The process of claim 1 wherein depleting of nutrient is to a concentration of 0.5 millimolar or less.

A3 6. (Amended) The process of claim 2 wherein the algal microorganism is selected from the group consisting of [?] a green, red, brown, and blue-green algae.

A4 10. (Amended) The process of claim 6 wherein the green algae is *Chlamydomonas reinhardtii*.

REMARKS

The Official Action and the cited references have been carefully reviewed. The review indicates that the claims, especially as amended, recites patentable subject matter and should be allowed. Reconsideration and allowance are therefore respectfully requested.

Before contending with the grounds upon which the rejections have been made, a brief summarization of the essentials of the reversible physiological process for temporal separation of hydrogen evolution and production of hydrogen in an algal microorganism will be provided to establish a clearer line of demarcation between the invention process and those processes disclosed in the cited and applied references.

In contemporary times where it is known that a substitute for fossil fuels includes hydrogen produceable from green algae, in which the reaction is catalyzed by reversible hydrogenase, and wherein the activity of the hydrogenase is immediately inactivated in the presence of light by photosynthetically generated O₂, applicants are the first to provide a sustainable process of photosynthetic hydrogen production in an algal culture that does not rely on mechanical means or chemical manipulations to cells. The process comprises:

- (a) growing a culture of the cells of the algal microorganism in a medium under illuminated conditions to accumulate an endogenous substrate;
- (b) depleting a nutrient selected from the group consisting of sulfur, iron, and/or manganese from the medium;
- (c) sealing the culture from atmospheric oxygen;
- (d) incubating the culture in light to provide a rate of light-induced oxygen production equal to or less than a rate of cellular respiration; and

(e) collecting an evolved gas that includes hydrogen.

Claims 1-9 were rejected as being anticipated by Weetall under 35 USC §102(b). *W 90:0 076*

Applicants respectfully traverse this rejection and request reconsideration for the reasons hereinafter provided.

A careful review of Weetall reveals that it is directed to a reactor for stabilized microbes having photometabolic activity. The reactor is used to produce hydrogen continuously by reacting a solution of a substrate in the presence of light with a photometabolically active microbe. The active microbe may be a blue-green algae that is stabilized by continuous biophotolysis of water by oxidizing the water and reducing NADP to NADPH.

*suggested
but not
emplified.*

On the other hand, and by contrast, the invention process affects a reversible physiological process for temporal separation of oxygen evolution and sustains photosynthetic hydrogen production in an algal microorganism by growing a culture of the microorganism under illuminated conditions to accumulate an endogenous substrate, depleting a nutrient such as sulfur from the medium, sealing the culture from atmospheric oxygen, incubating the culture in light to provide a rate of light-induced oxygen production equal to or less than the rate of cellular respiration, and collecting an evolved gas that includes hydrogen.

W 1076
No where in Weetall is there any reference to or mention of, depleting a nutrient in the algal microorganism in a culture medium, sealing the culture from atmospheric oxygen to avoid deactivation of hydrogenase in the presence of oxygen to provide a sustained production of hydrogen by virtue of incubating the culture in light to provide a rate of light-induced oxygen production equal to or less than the rate of cellular respiration to ensure sustained production of hydrogen by virtue of deactivation of hydrogenase.

*W 1076
uses
"sustained"
culture
(no
nutrients)*

Withdrawal of the rejection is respectfully requested.

Claims 1-8 and 10 were rejected as being anticipated by Greenbaum under 35 USC 102(b). *US 4442211*

Applicants respectfully traverse this rejection and request reconsideration for the reasons hereinafter provided.

A review of Greenbaum shows that its process produces hydrogen by subjecting algae in an aqueous phase to light irradiation; said process being more efficient than the prior art at the time of 1982 as a result of bleaching the cultured algae during a first period of irradiation in a culture medium in an aerobic atmosphere until it has regained color and then subjecting this algae to a second period of irradiation wherein hydrogen is produced at an enhanced rate.

Unlike the present invention, in Greenbaum, there is no acknowledgement of or reference to, the problem of the need for deactivation of hydrogenase in the presence of oxygen during photosynthetic hydrogen production in order to sustain the production of hydrogen.

Therefore, it is not surprising that no solution to this problem is even hinted at in Greenbaum.

On the other hand, applicants' invention as presently resolves the problem by depleting nutrients from the algal microorganism in a medium and affects temporal separation of oxygen evolution and incubates the culture in light to provide a rate of light-induced oxygen production less than the rate of cellular respiration to ensure sustained production of hydrogen. Inasmuch as Greenbaum lacks applicants' process steps of depleting a nutrient from the algal microorganism, temporal separation of the oxygen evolution and incubating the culture in light so as to provide a rate of light-induced oxygen production equal to or less

no mechanism is disclosed but same steps

✓ ?

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than the rate of cellular respiration to arrive at sustained hydrogen production, it is manifestly clear that Greenbaum fails to anticipate applicants' invention as presently recited.

Withdrawal of the rejection is respectfully requested.

Claims 1-10 were rejected as being unpatentable over Greenbaum and Weetall taken with Wykoff et al. and Melis Anastasios under 35 USC 103(a).

Greenbaum and Weetall have been discussed at length above; however, it is worthwhile reiterating that neither of these references address the problem of providing sustained production of hydrogen by avoiding deactivation of hydrogenase in the presence of oxygen by practicing applicants' invention as presently recited.

The deficiencies of these references are not compensated for by any teachings in the secondary references of Wykoff et al., and Melis Anastasios.

This is so because, Wykoff et al. merely pertains to disclosing the extent to which the light-saturated rate of photosynthetic O₂ evolution declines in *Chlamydomonas reinhardtii* upon P and S starvation. This publication makes no reference to or acknowledgement of the problem of not being able to sustain hydrogen production by virtue of the deactivation of hydrogenase in the presence of oxygen during the photosynthetic hydrogen production. Neither does Wykoff et al. provide any solution to this problem which it fails to appreciate or acknowledge.

A review of Melis shows that it is applicants' own publication published after the present application was filed. Applicants have incorporated herewith, an Affidavit Under 37 CFR §1.131 swearing back of this reference. Accordingly, this reference is not available against the present invention.

Even if the teachings in the secondary reference of Wykoff et al. were combined with the teachings of Greenbaum and Weetall, applicants' invention as presently recited would not ensue.

Neither would applicants' invention be rendered obvious. This is so because neither of the primary references address nor resolve the problem of providing sustained production of hydrogen by avoiding deactivation of hydrogenase in presence of oxygen. Therefore, even if the Wykoff et al. teachings of the extent to which the light saturated rate of photosynthetic O₂ evolution declines in *Chlamydomonas reinhardtii* upon P and S starvation were substituted into the processes of the primary references, such a substitution would clearly not provide a skilled person in the art with means for sustaining production of hydrogen by avoiding deactivation of hydrogenase in the presence of oxygen, as required by applicants' invention, as presently recited.

Withdrawal of the rejection is respectfully requested.

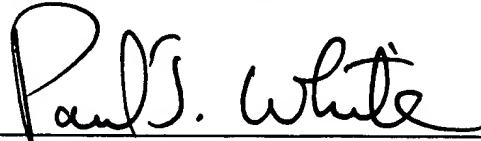
Note is taken of the rejections of claim 1-10 under the second paragraph of 35 USC 112 on grounds of indefiniteness; however, in view of the amendments made to the claims, these rejections are no longer applicable.

Note is taken of the objections raised to drawing Figures 1-7 under 37 CFR § 1.84 on grounds that the character of the lines, numbers and letters are not uniform and not plain or legible; however, in view of the formalized corrections of the characters and letters of Figures 1-7, and re-submission of these corrected and formalized figures, the objection is no longer applicable.

In view of the foregoing amendments, remarks, Affidavit under 37 CFR § 1.131, corrected drawing figures and arguments advanced, it is believed that the application is now in condition for allowance and early notification of the same is earnestly solicited.

Respectfully submitted,

Dated: May 7, 2002.

A handwritten signature in black ink, reading "Paul J. White". The signature is written in a cursive style with a horizontal line underneath.

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